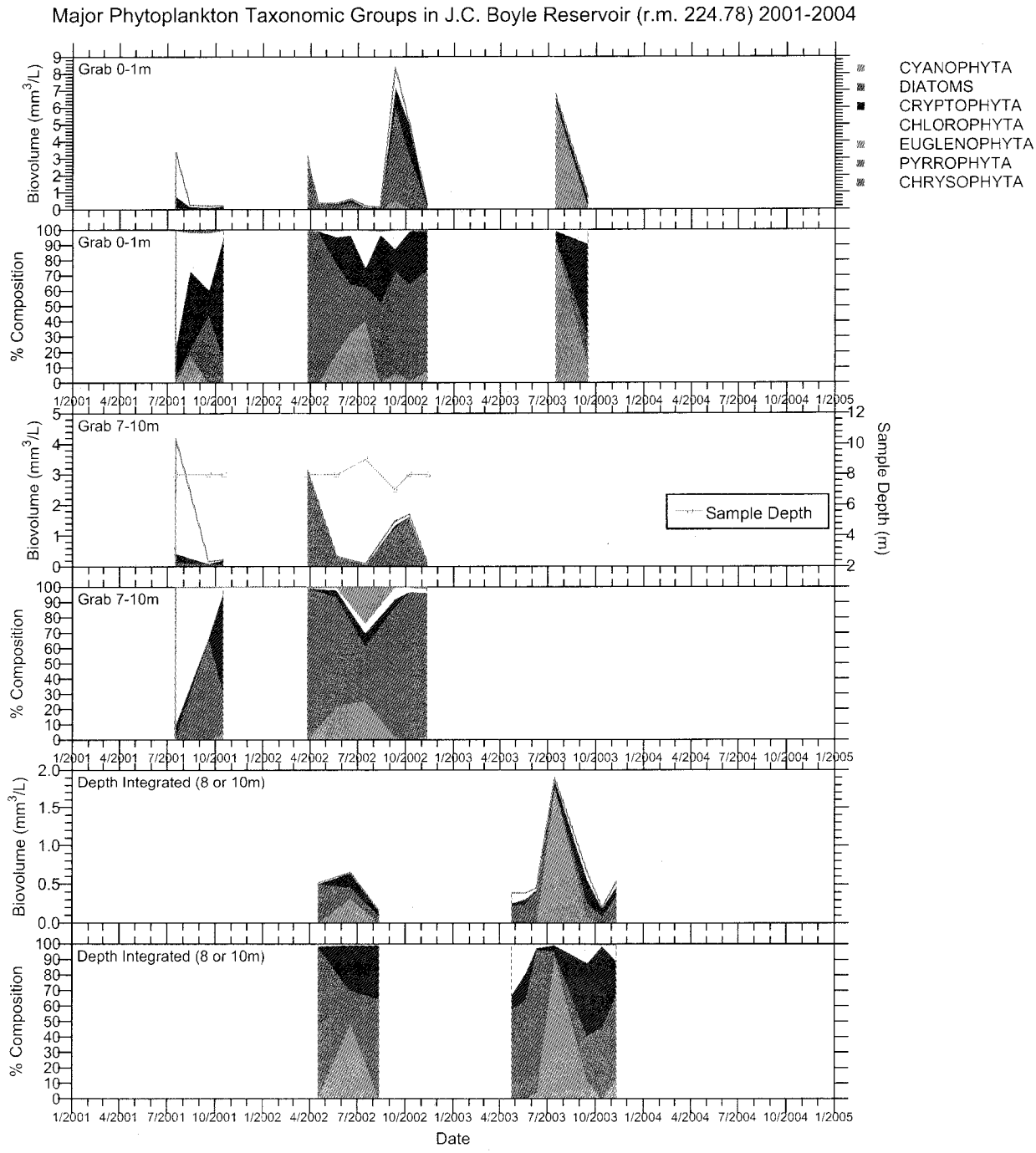

Figure 12. Biovolume and percent biovolume of major taxonomic groups of phytoplankton for samples collected in J.C. Boyle Reservoir in the years 2001-2003 (no samples collected in 2004). Each sampling depth is shown is a separate panel.

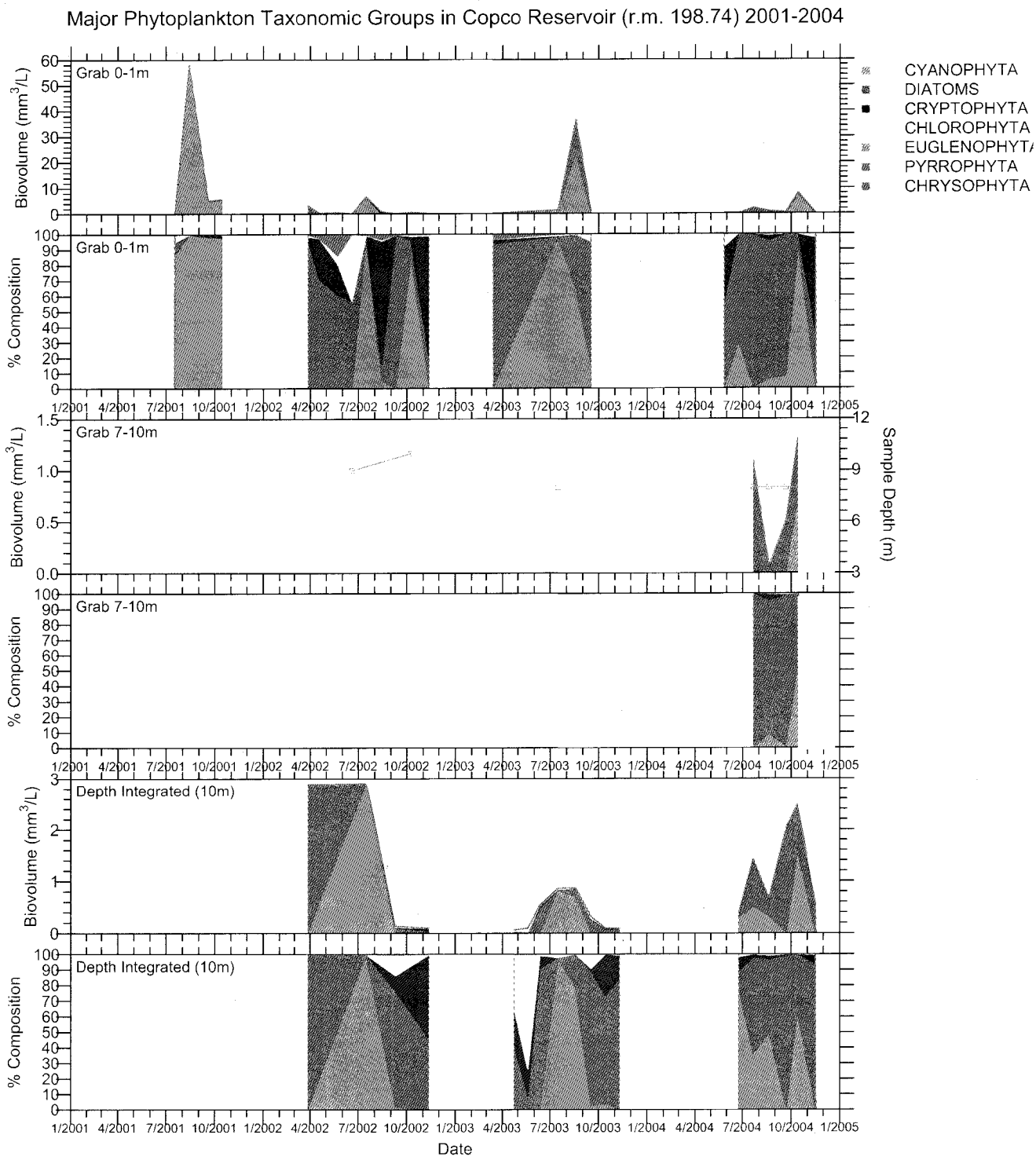

Figure 13. Biovolume and percent biovolume of major taxonomic groups of phytoplankton for samples collected in Copco Reservoir in the years 2001-2003 (no samples collected in 2004). Each sampling depth is shown is a separate panel. A few samples were also collected at depth greater than 10m, but those samples are not shown here.

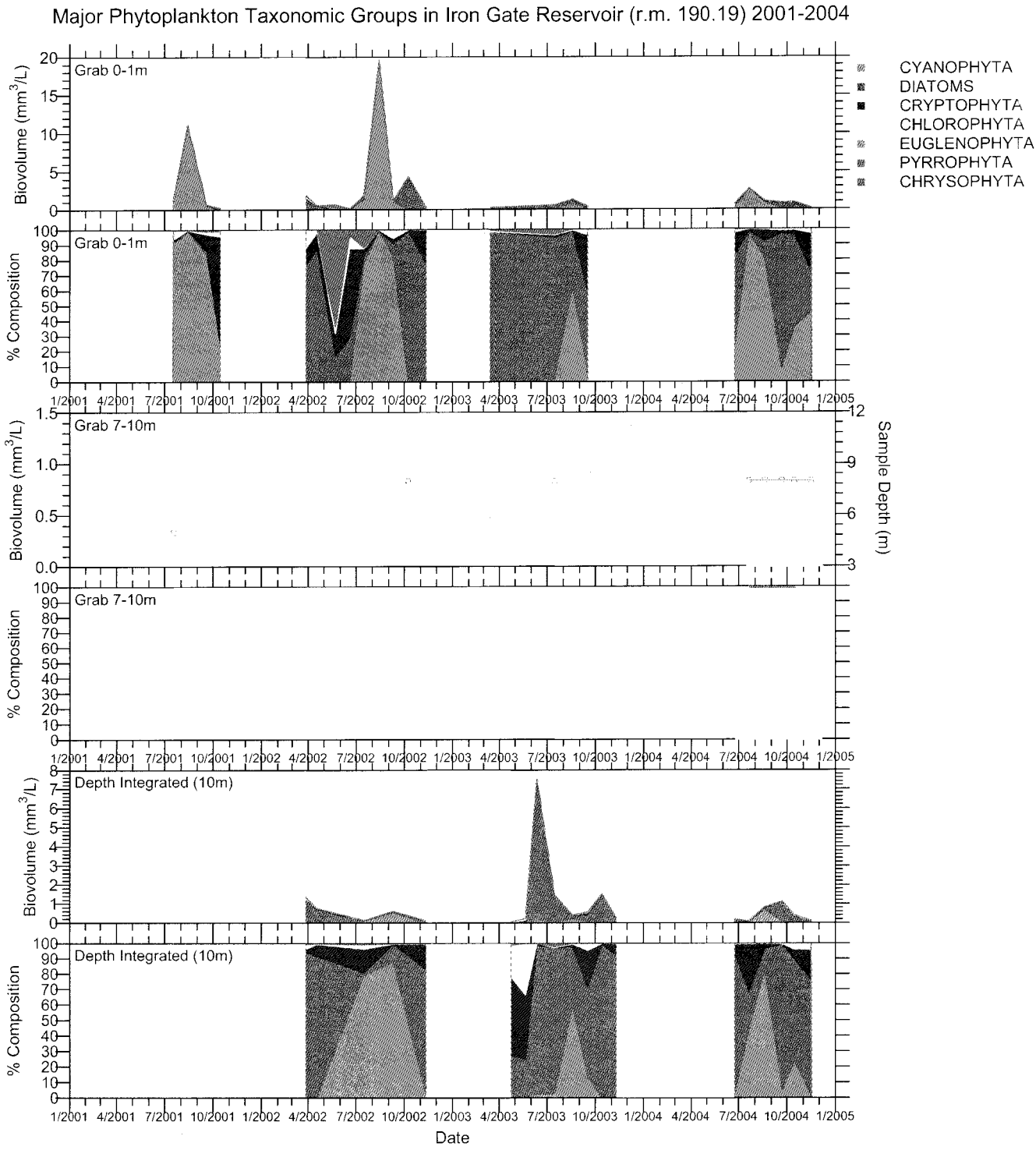

Figure 14. Biovolume and percent biovolume of major taxonomic groups of phytoplankton for samples collected in Iron Gate Reservoir in the years 2001-2003 (no samples collected in 2004). Each sampling depth is shown is a separate panel. A few samples were also collected at depth greater than 10m, but those samples are not shown here.

Species Composition

Seasonal trends for dominant species are shown for each station ordered longitudinally from UKL to the Klamath River above the Shasta River confluence in Figures 15-19. The intent of this section is not to provide detailed information on individual species, but rather to determine those species that comprise the major taxonomic groups described above in Figures 7-11. To improve graphical presentation, genera with more than one species observed were combined.

As discussed above, *Aphanizomenon* (APFA) is the major cyanophyte in water exiting UKL, often comprising >90% of the total Cyanophyta (Figure 15). There were also several incidences when *Microcystis aeruginosa* (MSAE) accounted for a small portion (<10%) of the cyanophyte biovolume leaving UKL. Moving downstream, APFA continues to dominate the Cyanophyta and MSAE is not noted again at considerable levels until Bel JC Boyle Dam (Figure 17). The seasonal APHA peak becomes more restricted as the river continues to the Keno and JC Boyle reaches (Figure 16), with increased summer dominance by *Cryptomonas* (e.g., CXER), *Cyclotella* , and *Melosira.* Spring and early-summer genera in this reach included the diatoms *Fragilaria*, *Stephanodiscus*, and *Asterionella.*

In the bypass reach Abv JCBPH (RM 220.4; Figure 17) where water is comprised mainly of spring inflow rather than reservoir or lake releases, the community was more diverse, and tended to be dominated by periphytic or attached diatom genera such as *Diatoma*, *Cocconeis*, *Gomphonema*, *Navicula*, and *Nitzschia.* Although this trend in increased periphytic diatoms continued to the Above Copco station (Abv Copco; RM 206.42), APFA again increased as water released from the JC Boyle Powerhouse mixed with water from the bypass reach.

Periphytic species then declined substantially and were replaced by more planktonic species in the Copco Iron Gate Reservoir complex (Figure 18). For example, APFA increased from 20.1% of the June-September biovolume above Copco to 70.8% of the biovolume in Copco Reservoir (Tables 4 and 5). Likewise, the second most dominant species in Copco reservoir for this period was MSAE (14.3%), a species that did not rank in the top ten dominant species above Copco, and was present at levels≤5% at upstream stations. Other planktonic genera that were periodically important in Copco Reservoir include the cyanophyte *Anabaena,* and the diatoms *Fragilaria* and *Stephanodiscus* (Figure 18). Below Copco, APFA decreased to 30.4% (RM 196.45; Table 4) and the period of APFA dominance was more restricted (Figure 18). In addition, diatoms increased in importance with *Nitzschia* and *Fragilaria* accounting for 32.4% of the biovolume on a seasonal basis (Table 4). In Iron Gate Reservoir, APFA again increased to 83% of the biovolume, with *Gloeotrichia* (GTEC), another nitrogen-fixing blue-green, ranking second at 5.7% (Table 4). MSAE decreased in importance in Iron Gate Reservoir, but did show annual peaks in most years at the site below Iron Gate Dam (RM 189.73) where MSAE comprised 16.8% of biovolume in August 2004 and 6.7% in September 2004.

Seasonal patterns in the reservoirs generally included spring dominance by diatoms (e.g., *Stephanodiscus* and *Melosira*), summer dominance by the Cyanophyta (e.g., *Aphanizomenon*, *Microcystis,* and *Gloeotrichia*), and fall dominance by *Cryptomonas* and *Fragilaria* (Figure 18). Although limited by sampling frequency, there is an indication that 14-17 miles below Iron Gate Dam at both RM 176 and RM 173 that the periphytic diatom *Cocconeis* increases substantially in importance (Figure 18; Table 4).

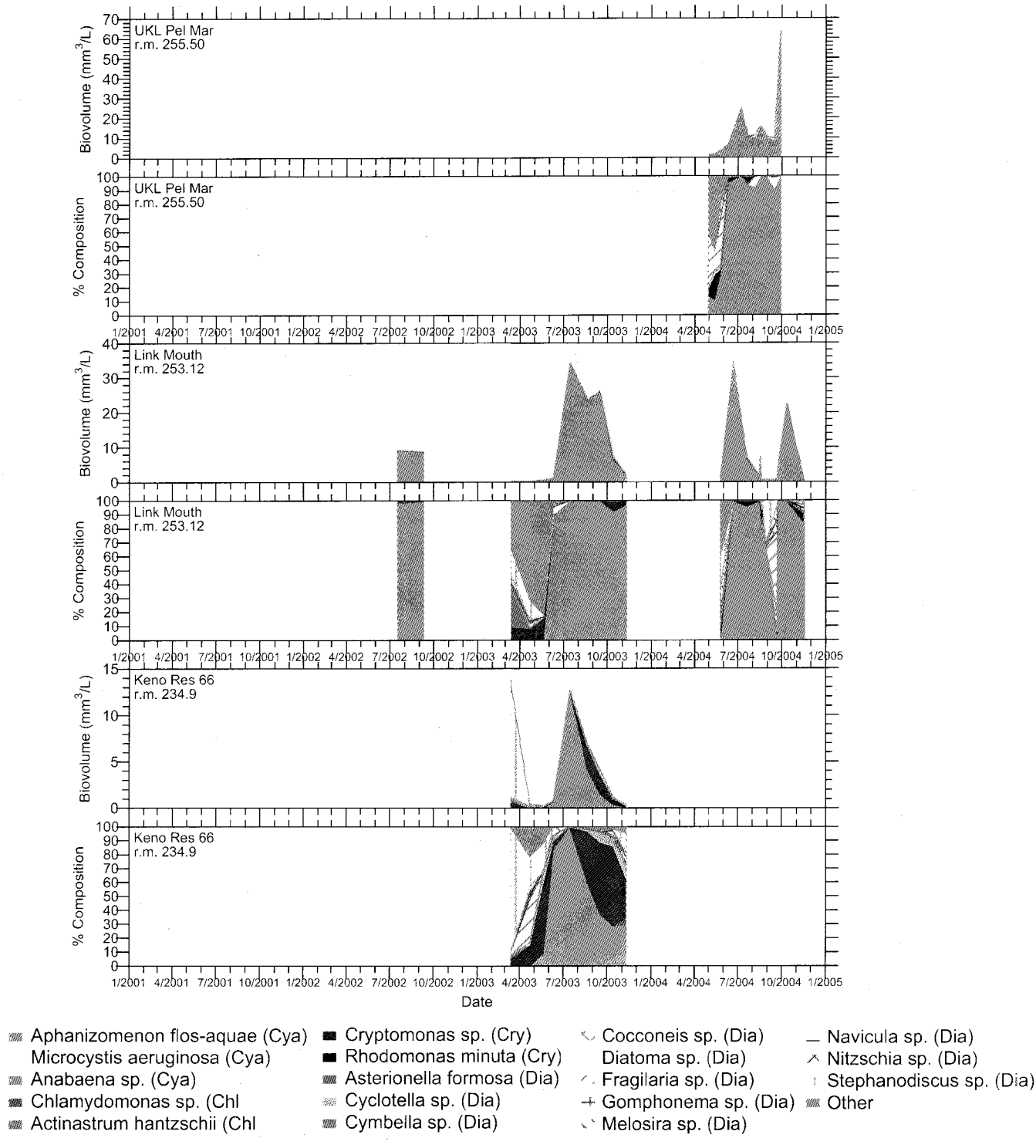

Figure 15. Biovolume and percent biovolume of dominant species of phytoplankton for surface samples collected in the years 2001-2004. Sites are listed in downstream order: Upper Klamath Lake at Pelican Marina, Link River at its mouth, and Keno Reservoir at the Highway 66 Bridge.

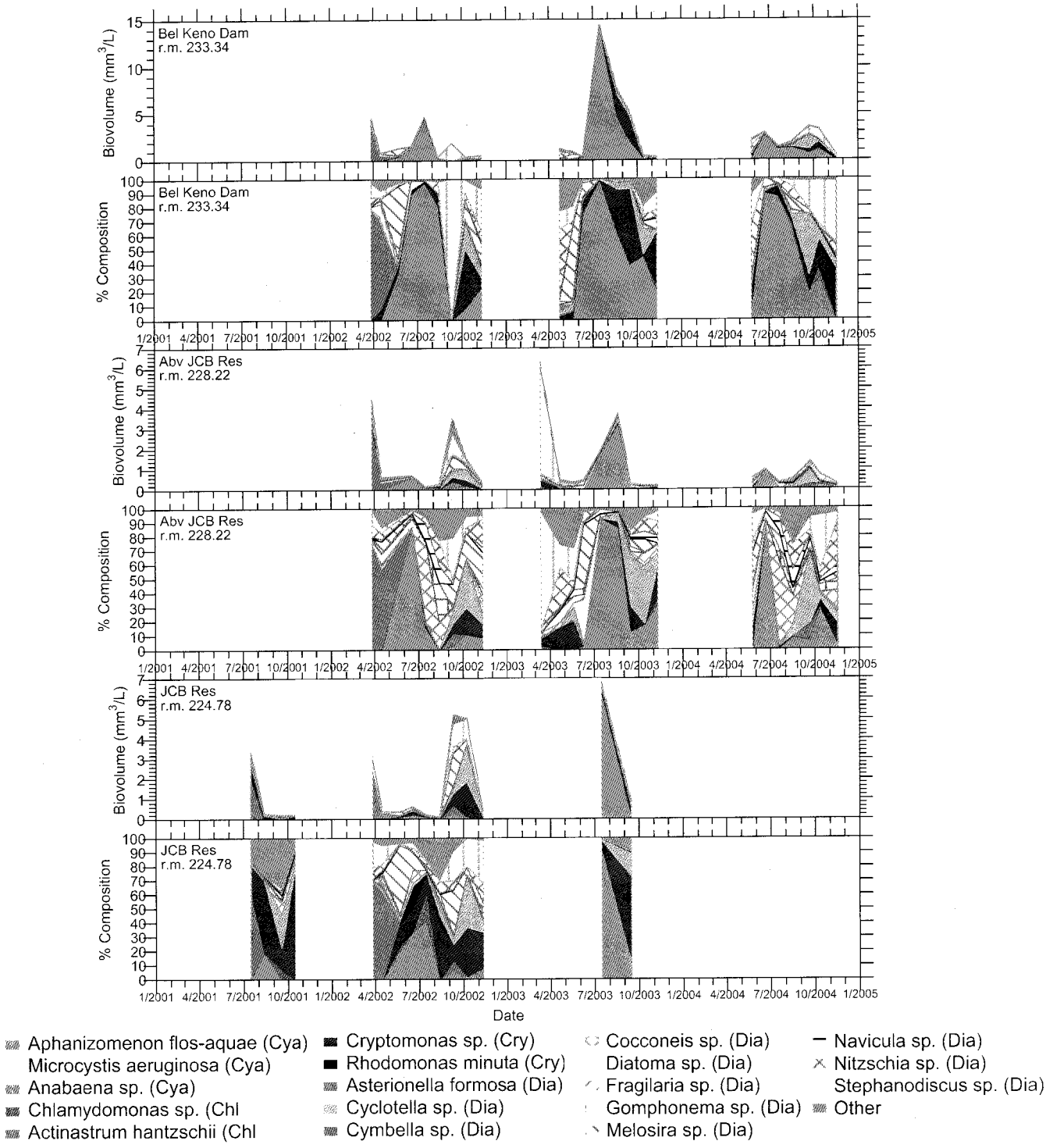

Figure 16. Biovolume and percent biovolume of dominant species of phytoplankton for surface samples collected in the years 2001-2004. Sites are listed in downstream order: Keno Dam, Above J.C. Boyle Reservoir, and J.C. Boyle Reservoir.

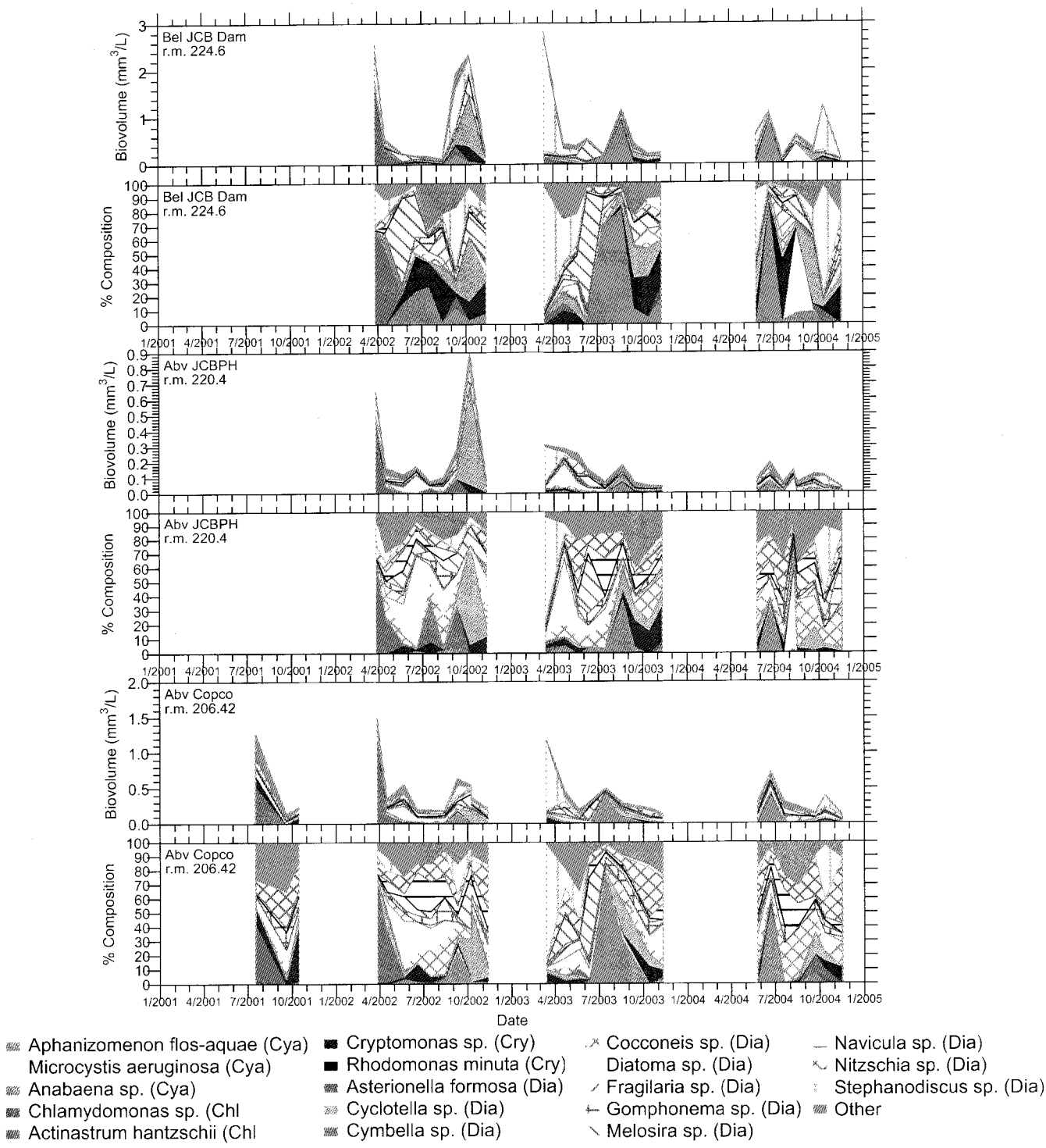

Figure 17. Biovolume and percent biovolume of dominant species of phytoplankton for surface samples collected in the years 2001-2004. Sites are listed in downstream order: Below J.C. Boyle Dam, Above J.C. Boyle Powerhouse, and Above Copco Reservoir.

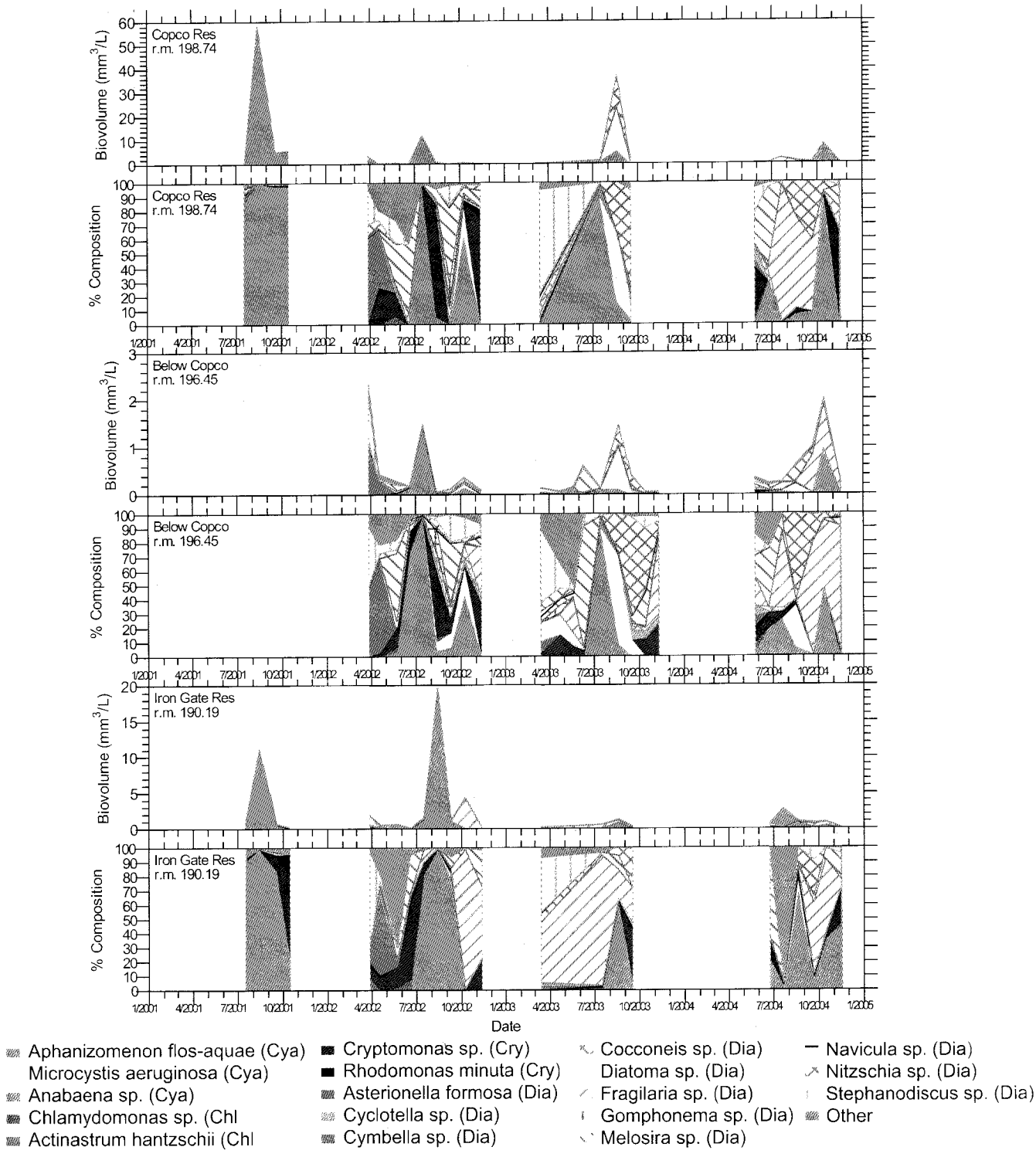

Figure 18. Biovolume and percent biovolume of dominant species of phytoplankton for surface samples collected in the years 2001-2004. Sites are listed in downstream order: Copco Reservoir, Below Copco Dam, and Iron Gate Reservoir.

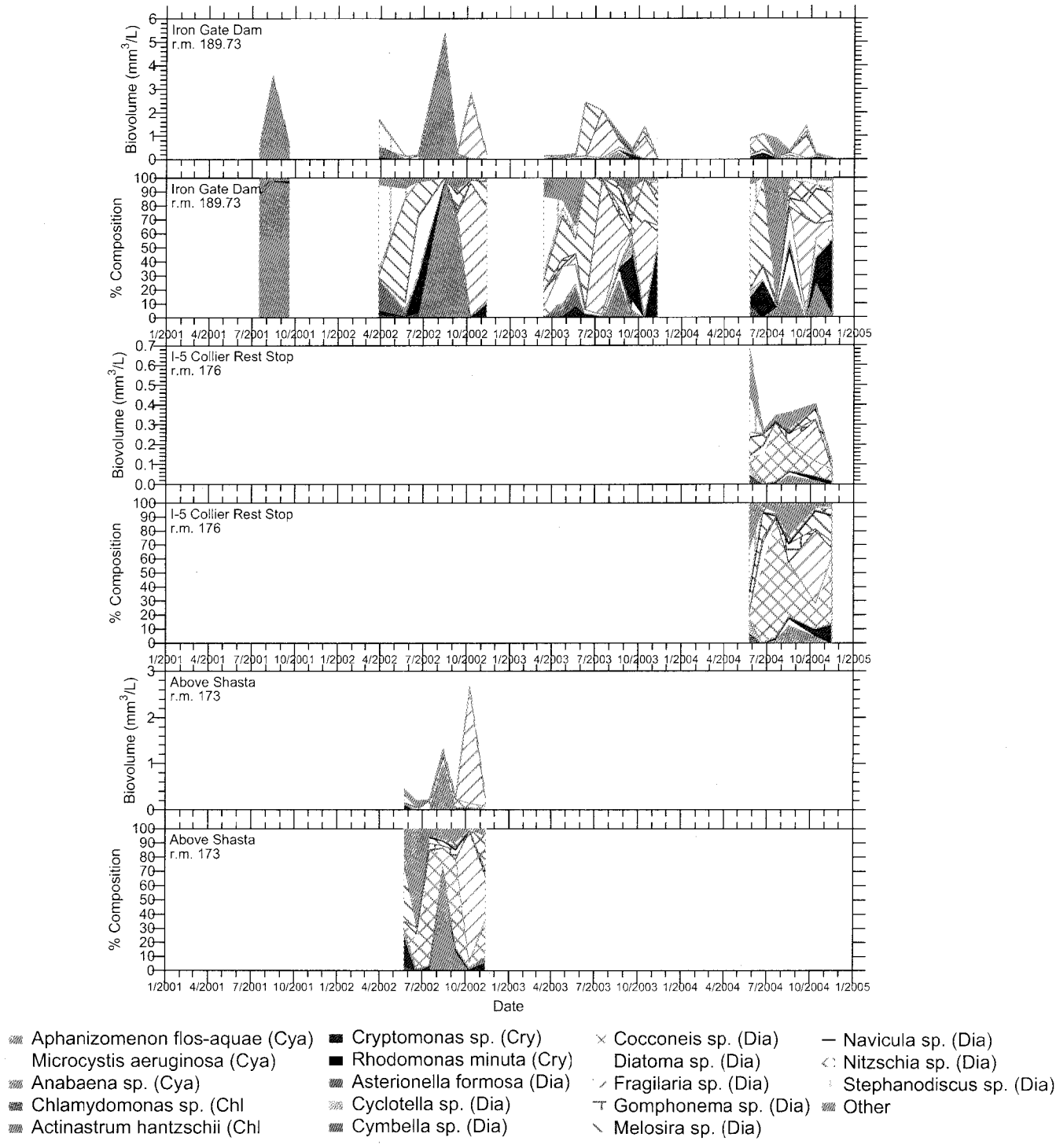

Figure 19. Biovolume and percent biovolume of dominant species of phytoplankton for surface samples collected in the years 2001-2004. Sites are listed in downstream order: Iron Gate Dam, Above the Shasta River, at the Interstate 5 Collier Rest Area.

Table 4. Biovolume for 10 most dominant species at each site, for surface samples collected in the years 2001-2004, June 1 – Sept 30. See Table 5 for a key to four-letter species codes.

River Mile	Site Name	N		Species ranked by mean biovolume (mm^3/L) for samples June 1 - Sept. 30										
				All	1	2	3	4	5	6	7	8	9	10
173/176	I-5 Shasta	7	Species		COPC	APFA	SNUL	EPSX	GFSB	DTVL	MLVR	OSXX	MLGR	RHCU
			Mean	0.4	0.1578	0.1519	0.0220	0.0188	0.0110	0.0101	0.0080	0.0068	0.0058	0.0048
			Percent		36.8%	35.4%	5.1%	4.4%	2.6%	2.4%	1.9%	1.6%	1.4%	1.1%
189.73	IG Dam	15	Species		APFA	FRCR	MLGR	GTEC	CXER	DTVL	NZPL	MLGA	COPC	MSAE
			Mean	1.4	0.7154	0.2200	0.2059	0.0501	0.0426	0.0248	0.0237	0.0185	0.0171	0.0112
			Percent		50.8%	15.6%	14.6%	3.6%	3.0%	1.8%	1.7%	1.3%	1.2%	0.8%
190.19	IG Res	15	Species		APFA	GTEC	FRCR	CXER	MLGR	NZPL	CJHR	MSAE	RDMN	MLGA
			Mean	3.0	2.4966	0.1724	0.0918	0.0456	0.0391	0.0322	0.0270	0.0080	0.0072	0.0049
			Percent		83.0%	5.7%	3.0%	1.5%	1.3%	1.1%	0.9%	0.3%	0.2%	0.2%
196.45	Bel Copco	12	Species		APFA	NZPL	MSAE	FRCR	MLAM	MLGR	MLGA	CXER	EDEL	STBI
			Mean	0.5	0.1653	0.0994	0.0941	0.0767	0.0261	0.0193	0.0144	0.0109	0.0044	0.0040
			Percent		30.4%	18.3%	17.3%	14.1%	4.8%	3.6%	2.7%	2.0%	0.8%	0.7%
198.74	Copco Res	16	Species		APFA	MSAE	NZPL	FRCR	CXER	CHXX	MLGR	CJHR	RDMN	COPC
			Mean	7.9	5.6065	1.1307	0.8561	0.2084	0.0538	0.0083	0.0069	0.0047	0.0040	0.0035
			Percent		70.8%	14.3%	10.8%	2.6%	0.7%	0.1%	0.1%	0.1%	0.1%	0.0%
206.42	Abv Copco	14	Species		APFA	ATHN	COPC	NVTP	DTVL	NZDS	CXER	RHCU	NZFR	STHN
			Mean	0.4	0.0763	0.0373	0.0320	0.0223	0.0207	0.0168	0.0166	0.0116	0.0105	0.0104
			Percent		20.1%	9.8%	8.4%	5.9%	5.5%	4.4%	4.4%	3.1%	2.8%	2.7%
220.40	Abv JCB PH	13	Species		DTVL	APFA	COPC	MLAM	MSAE	ATHN	NVTP	RHCU	CCMG	SNUL
			Mean	0.1	0.0146	0.0138	0.0112	0.0074	0.0065	0.0062	0.0059	0.0058	0.0054	0.0043
			Percent		11.4%	10.7%	8.7%	5.8%	5.0%	4.8%	4.6%	4.5%	4.2%	3.3%
224.60	Bel JCB Dam	12	Species		APFA	STHN	CCMG	MLAM	MSAE	CXER	MLGR	SNUL	SCQD	ATHN
			Mean	0.6	0.2092	0.0713	0.0402	0.0359	0.0307	0.0273	0.0217	0.0162	0.0154	0.0136
			Percent		36.6%	12.5%	7.0%	6.3%	5.4%	4.8%	3.8%	2.8%	2.7%	2.4%
224.78	JCB Res	9	Species		APFA	ATHN	CXER	STHN	MLAM	MLGR	CCMG	SCQD	SCAC	SFSR
			Mean	2.0	0.8004	0.2681	0.2570	0.1205	0.1138	0.0814	0.0619	0.0283	0.0257	0.0231
			Percent		40.7%	13.6%	13.1%	6.1%	5.8%	4.1%	3.2%	1.4%	1.3%	1.2%
228.22	Abv JCB Res	12	Species		APFA	CCMG	STHN	SNUL	MLAM	COPC	CXER	ATHN	MLGR	NVCV
			Mean	1.2	0.5575	0.1097	0.0922	0.0568	0.0452	0.0437	0.0369	0.0366	0.0277	0.0148
			Percent		47.2%	9.3%	7.8%	4.8%	3.8%	3.7%	3.1%	3.1%	2.3%	1.3%
233.34	Keno Dam	12	Species		APFA	CXER	STHN	CCMG	STBI	MLGR	NZPL	MLGA	RDMN	NZAC
			Mean	3.8	2.7610	0.4814	0.1934	0.1771	0.0258	0.0200	0.0163	0.0146	0.0102	0.0096
			Percent		72.5%	12.6%	5.1%	4.7%	0.7%	0.5%	0.4%	0.4%	0.3%	0.3%
253.12	Link Mouth	19	Species		APFA	FRCR	CXER	COPC	STAS	NZPL	GFSB	MLGR	STAM	DTVL
			Mean	8.6	8.5183	0.0224	0.0206	0.0091	0.0089	0.0086	0.0075	0.0057	0.0037	0.0029
			Percent		98.7%	0.3%	0.2%	0.1%	0.1%	0.1%	0.1%	0.1%	0.0%	0.0%
255.50	UKL Pel Mar	8	Species		APFA	MSAE	CXER	CHXX	CXOV	RDMN	KMXX	GDXX	MLGR	MGXX
			Mean	19.1	18.7380	0.2018	0.0580	0.0303	0.0182	0.0129	0.0120	0.0069	0.0058	0.0056
			Percent		98.1%	1.1%	0.3%	0.2%	0.1%	0.1%	0.1%	0.0%	0.0%	0.0%

Table 5. Key to four-letter species codes for most dominant phytoplankton species at sites listed in Table 4 for surface samples in years 2001-2004, with species listed in alphabetical order.

Species Code	Species Name	Major Taxonomic Group
APFA	*Aphanizomenon flos-aquae*	Cyanophyta
ATHN	*Actinastrum hantzschii*	Chlorophyta
CCMG	*Cyclotella meneghiniana*	Diatoms
CHXX	*Chlamydomonas sp*	Chlorophyta
CJHR	*Ceratium hirundinella*	Pyrrophyta
COPC	*Cocconeis placentula*	Diatoms
CXER	*Cryptomonas erosa*	Cryptophyta
CXOV	*Cryptomonas ovata*	Cryptophyta
DTVL	*Diatoma vulgare*	Diatoms
EDEL	*Eudorina elegans*	Chlorophyta
EPSX	*Epithemia sorex*	Diatoms
FRCR	*Fragilaria crotonensis*	Diatoms
GDXX	*Glenodinium sp*	Pyrrophyta
GFSB	*Gomphonema subclavatum*	Diatoms
GTEC	*Gloeotrichia echinulata*	Cyanophyta
KMXX	*Chromulina sp*	Chrysophyta
MGXX	*Mougeotia sp*	Chlorophyta
MLAM	*Melosira ambigua*	Diatoms
MLGA	*Melosira granulata angustissima*	Diatoms
MLGR	*Melosira granulata*	Diatoms
MLVR	*Melosira varians*	Diatoms
MSAE	*Microcystis aeruginosa*	Cyanophyta
NVCV	*Navicula cryptocephala veneta*	Diatoms
NVTP	*Navicula tripunctata*	Diatoms
NZAC	*Nitzschia acicularis*	Diatoms
NZDS	*Nitzschia dissipata*	Diatoms
NZFR	*Nitzschia frustulum*	Diatoms
NZPL	*Nitzschia palea*	Diatoms
OSXX	*Oscillatoria sp*	Cyanophyta
RDMN	*Rhodomonas minuta*	Cryptophyta
RHCU	*Rhoicosphenia curvata*	Diatoms
SCAC	*Scenedesmus acuminatus*	Chlorophyta
SCQD	*Scenedesmus quadricauda*	Chlorophyta
SFSR	*Sphaerocystis schroeteri*	Chlorophyta
SNUL	*Synedra ulna*	Diatoms
STAM	*Stephanodiscus astraea minutula*	Diatoms
STAS	*Stephanodiscus astraea*	Diatoms
STBI	*Stephanodiscus binderanus*	Diatoms
STHN	*Stephanodiscus hantzschii*	Diatoms

The overall longitudinal trend for individual species for the June-September period showed a declining trend in APFA from UKL to above Copco and a subsequent increase in Copco and Iron Gate Reservoirs (Table 4). As expected based on the trend in both nitrogen-fixing phytoplankton (NFP) and Cyanophyta described above, 85.1% of the biovolume in Copco reservoir surface stations was comprised of APFA and MSAE, and 88.7% was comprised of APFA and GTEC in Iron Gate Reservoir.

SUMMARY AND CONCLUSIONS

The purpose of this report was to provide an analysis of PacifiCorp's 2001-2004 phytoplankton dataset for the Klamath River from Upper Klamath Lake to above the Shasta River's confluence with the Klamath. Due to the importance of Upper Klamath Lake (UKL) to the Klamath River's nutrient and algal dynamics, phytoplankton data collected by the Klamath Tribes near the outlet of UKL were also used to provide a context for comparison.

Longitudinal trends for two time periods (all available sample dates and, then, June-September) showed the same basic trend of decreasing total phytoplankton biovolume, decreasing nitrogen-fixing phytoplankton (NFP) biovolume, and decreasing NFP percent composition from UKL to the station above Copco Reservoir. Downstream from this station these trends reverse as the river continues through the impounded areas of Copco and Iron Gate reservoirs, with total biovolume, NFP biovolume, and percent NFP all then increasing substantially, especially during the June-September period. The most pronounced change occurred in the upper 25th percentile (upper quartile) of the distribution, where compared to an NFP composition of 24.5% above Copco, reservoir NFP percent composition values returned to levels closer to those of UKL, exceeding 90% in both reservoirs. Thus, despite a decline in the downstream magnitude along the longitudinal profile between UKL and above Copco Reservoir, all parameters (total biovolume, NFP biovolume, and NFP percent biovolume) showed clear increases (by tens to hundreds of times) in the Copco/Iron Gate Reservoir complex during the June-September period.

Although not as pronounced as for the analyses of phytoplankton biovolume, the longitudinal trend in chlorophyll *a*, which provides an approximation of algal biomass, showed the same basic decreasing trend between UKL and above Copco Reservoir, and then increased (both median and upper quartile values) as the river traveled through the reservoir complex.

As expected as the system changed from the lacustrine environment of UKL to the riverine environment of the Klamath River, diatoms increased in prevalence downstream before decreasing again in the Copco/Iron Gate Reservoir complex as the Cyanophyta again dominated. Moreover, because Cyanophyta (blue-green algae) are comprised chiefly of algae from the NFP group, the longitudinal trend in both total biovolume and percent biovolume of the Cyanophyta was similar to that of NFP. The trend in Cyanophyta percent composition was more pronounced through the reservoir complex than absolute biomass, with levels in Copco and Iron Gate increasing from 5% above Copco to 50% and 82% in Copco and Iron Gate Reservoirs, respectively. Earlier analyses by Kann (2006) showed this same trend for *Microcystis aeruginosa,* a species that does not fix nitrogen, but that is a member of the Cyanophyta.

Consistently more pronounced trends in the upper quartile (UQ) relative to the median indicate that periodic high values or bloom events of NFP and Cyanophyta occurred in the reservoir complex relative to stations directly upstream.

Periodic multiple depth sampling in JC Boyle, Copco, and Iron Gate Reservoirs from 2001-2004 tend to indicate that the composition of diatoms increases relative to the surface samples, and as expected, based on the dilution of surface water where algae tend to be more concentrated, overall water column biovolume was substantially lower than surface samples in all three reservoirs. Although overall biovolume was lower in the depth-integrated samples, they showed a seasonal pattern of blue-green dominance similar to the surface samples. These samples indicate the buoyant nature of most Cyanophyta which tend to be concentrated near the surface. Thus, the Iron Gate Dam station (RM 189.73) which consists of water drawn from ~30-40 ft at the outlet of Iron Gate Reservoir, showed reduced composition of Cyanophyta, although relative to Abv. Copco, the levels were higher and the period of dominance was prolonged.

Aphanizomenon (APFA) was the major cyanophyte leaving UKL, decreasing in importance downstream to the bypass reach where the community was more diverse and tended to be dominated by periphytic or attached diatom genera such as *Diatoma*, *Cocconeis*, *Gomphonema*, *Navicula*, and *Nitzschia*. Periphytic species then declined substantially and were replaced by more planktonic species in the Copco/Iron Gate Reservoir complex with APFA increasing from 20.1% of the June-September biovolume above Copco to 70.8% of the biovolume in Copco Reservoir. The second most dominant species in Copco reservoir for this period was *Microcystis aeruginosa* (MSAE, 14.3%), a species that did not rank in the top ten dominant species above Copco, and was present at levels ≤5% at stations directly upstream. In Iron Gate Reservoir, APFA again increased to 83% of the biovolume, with *Gloeotrichia* (GTEC), another nitrogen-fixing blue-green, ranking second at 5.7%. MSAE decreased in importance in Iron Gate Reservoir, but did show annual peaks in most years at the site below Iron Gate Dam (RM 189.73) where MSAE comprised 16.8% of biovolume in August 2004 and 6.7% in September 2004.

The overall longitudinal trend for phytoplankton biovolume and important nitrogen-fixing and bloom forming species all confirm the same declining trend from UKL to above Copco Reservoir, with a subsequent increase in the Copco/Iron Gate Reservoir complex.

These results are similar to the analysis of the PacifiCorp dataset for MSAE only (Kann 2006), where analyses showed increased incidence and magnitude of MSAE in Copco and Iron Gate Reservoirs relative to stations upstream. Although MSAE is not a nitrogen–fixing species, it is an important member of the Cyanophyta. As stated in Kann (2006) and Kann and Corum (2006) this trend of increasing Cyanophyta is consistent with literature showing that MSAE and other buoyant cyanobacteria such as APFA do not dominate in river conditions of turbulent mixing such as that known to occur in the Klamath River above Copco and Iron Gate Reservoirs. Because cyanophytes tend to thrive at low turbulent diffusivity (calm-stable conditions) when their flotation velocity exceeds the rate of turbulent mixing, they are favored in lake and reservoir environments that tend to be warmer and less turbulent than riverine ones (Reynolds 1986).

In conclusion, these analyses show that although the Klamath River receives a large loading of algal biomass (made up largely of the cyanophyte, APHA) from UKL, the analyzed data provide clear evidence that Copco and Iron Gate Reservoirs are providing habitat conditions that foster increased overall phytoplankton biovolume comprised largely of nitrogen-fixing cyanophyte species as well as

toxigenic MSAE. The relative increase in nitrogen-fixing species is important ecologically because these species have the potential to introduce additional nitrogen into the Klamath River system. Although nitrogen increases from algal fixation are not separable from reservoir sediment loading of nitrogen, nutrient budgets in Copco and Iron Gate Reservoirs for the year 2002 indicated that the reservoirs can act as both sources and sinks for nitrogen during the algal growing season (Kann and Asarian 2005).

Thus, this analysis underscores the need to account for reservoir hydrologic and nutrient alterations that foster increased phytoplankton growth in the Klamath River. Evaluation of impaired water quality due to hydrologic alterations attributable to the Klamath Hydroelectric Project should include such reservoir alterations as increased water retention time, higher water column stability, and internal sediment loading of nutrients. Although upstream loading of nutrients and algae are important, alterations to the river such as these can contribute to a further increase in algal production, especially of blue-green algal blooms.

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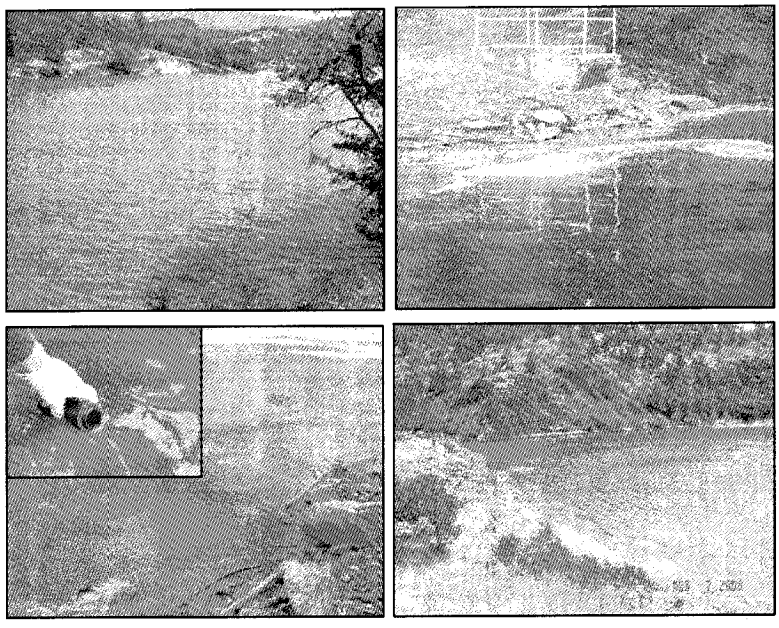
ELECTRONIC APPENDICES ON CD

A. Spreadsheet of PacifiCorp 2001-2004 phytoplankton data.

Aquatic Ecosystem Sciences LLC

TECHNICAL MEMORANDUM

Partial Seasonal Summary of 2006 Toxic *Microcystis aeruginosa* Trends in Copco and Iron Gate Reservoirs and the Klamath River, CA



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November, 2006

INTRODUCTION

As outlined in Kann and Corum (2006), Copco and Iron Gate Reservoirs (the lowermost projects of PacifiCorp's Klamath Hydropower Project-- KHP) experienced extensive blooms of toxigenic *Microcystis aeruginosa* (MSAE) in 2004 and 2005. These blooms were associated with high levels of microcystin, a potent hepatotoxin capable of causing chronic liver damage and acting as a tumor promoter (Carmichael 1995; Chorus et al. 1999; Chorus 2001).

The results of the 2005 sampling program demonstrated widespread and high abundance of toxigenic MSAE blooms in Copco and Iron Gate reservoirs from July-October, exceeding World Health Organization Moderate Probability of Adverse Health Effect Levels (WHO MPHAEL) for both cell density and toxin by 10 to over 1000 times Although both cell density and toxin data indicated that MSAE cells and microcystin were not detectable in the Klamath River directly above the reservoirs, detectable levels of both parameters were found directly below the reservoirs in 2005.

A similar toxic algae monitoring program was undertaken by the Karuk Tribe in 2006, and although microcystin toxin results are only available through September (October-November results are pending), the following technical memorandum serves a partial seasonal summary for 2006.

METHODS

During the 2006 sampling season, MSAE cell density, cell biovolume, and microcystin toxin samples were collected from a variety of shoreline and open-water sites, including standard open-water locations (Table 1 and Figure 1; Stations IR01, IR03, and CR01) and shoreline stations specifically sampled to assess the extent of toxic MSAE in the vicinity of public recreational access points (Figure 1).

Table 1. Phytoplankton/microcystin sampling locations in Copco and Iron Gate Reservoirs and Klamath River stations, 2006.

STATION NAME	STATION LAT/LON	Station Description	Shoreline (SL) or Open Water (OW)
CR01	N41 58.932 W122 19.694	Copco Res. Near Dam	OW
CRCC	N41 59.035 W122 19.802	Copco Res. Copco Cove Boat Ramp/Recreation Area	SL
CRMC	N41 58.441 W122 17.869	Copco Res. Mallard Cove Boat Ramp/Recreation Area	SL
IR01	N41 56.330 W122 25.930	Iron Gate Res. Near Dam	OW
IR03	N41 57.876 W122 25.389	Iron Gate Res. Upper 1/2	OW
IRCC	N41 58.368 W122 26.114	Iron Gate Res. Camp Creek Boat Ramp/Recreation Area	SL
IRJW	N41 57.721 W122 26.425	Iron Gate Res. Jay Williams Boat Ramp/Recreation Area	SL
KRAC	N41 58.345 W122 12.101	Klamath River Above Copco Reservoir	River
KRBI	N41 55.865 W122 26.532	Klamath River Below Iron Gate Reservoir	River
SV	N41 50.561 W 123 13.132	Seiad Valley	River
OR	N 41 18.336 W 123 31.895	Orleans	River

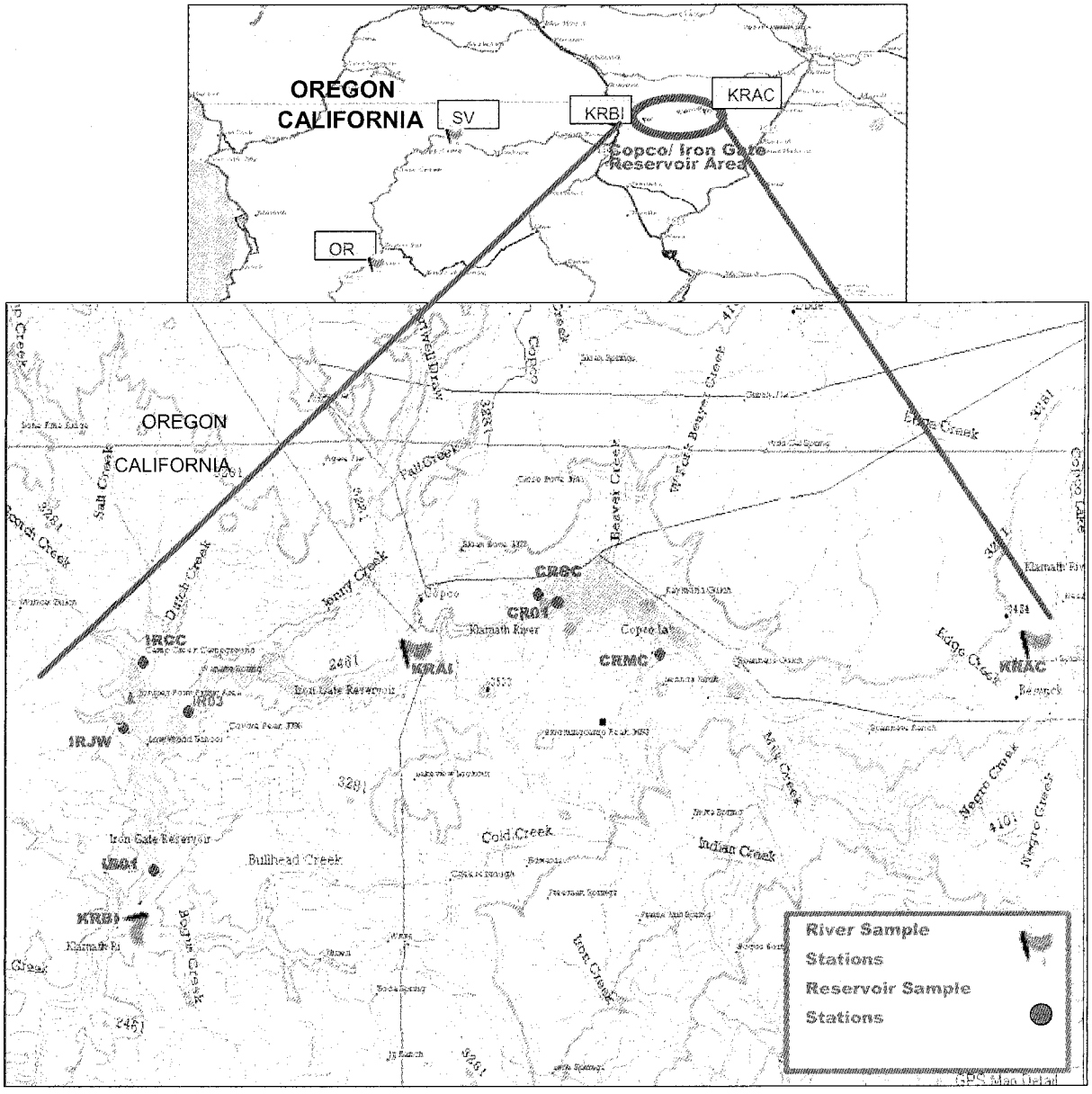


Figure 1. Location of Copco and Iron Gate Reservoir and Klamath River toxic cyanobacteria sampling stations, 2006.

The stations KRAC, KRBI, SV, and OR are Klamath River stations above Copco (KRAC) and Below Iron Gate (KRBI, SV, and OR) (Figure 1).

Shoreline and open-water samples taken at the surface consisted of grab samples of surface algal material, and both open-water samples taken at 1 m and samples collected at river stations KRAC and KRAI were taken with a Van-Dorn water collection bottle (KRAI data have not yet been received from the laboratory). Samples for microscopic determination of phytoplankton density and biovolume were preserved in Lugol's Iodine and sent to Aquatic Analysts in White Salmon, WA where enumeration and biovolume measurements are determined according to APHA Standard Methods (1992). Phytoplankton laboratory reports are contained in Electronic Appendix E1.

Samples for determination of microcystin toxin were placed in a cooler with gel-ice and shipped overnight air to Wright State University in Dayton, OH (CyanoHab Services Lab of Dr. Wayne Carmichael). These samples were analyzed for microcystin toxin using ELISA methodology (microcystin laboratory reports and methodology are contained in Electronic Appendix E2).

Cell density and toxin concentration were compared to WHO Moderate Probability of Adverse Health Effect thresholds (MPAHEL) for recreational waters as published in documents for the WHO and EPA (Falconer el al. 1999; Chorus and Cavalieri 2000). The MPAHEL is 100,000 cells/ml or 20 µg/L microcystin in the top 4 meters of surface waters, and the tolerable daily intake (TDI) is as described in (WHO 1998). The WHO (Falconer et al. 1999) further lists cyanobacterial scums in swimming areas as having a high probability of adverse health effects (i.e., the potential to cause acute poisoning) and recommends immediate action to prevent contact with scums. In addition, cell density and toxin concentration were also compared to levels used by the State of Oregon and adopted by the Posting Subcommittee of the Klamath Basin Blue-Green algae Working Group. These levels are 40,000 cells/ml of MSAE and 8 µg/L of microcystin. These levels are specific for MSAE and microcystin, whereas the WHO levels are general levels for a variety of toxigenic cyanobacteria.

RESULTS

Similar to the timing in 2005, the first visual detection of a cyanobacterial bloom in the reservoir system in 2006 was noted during regular biweekly sampling on July 12th and 13th at station CRCC in Copco Reservoir. Subsequent analyses showed >11 million MSAE cells per ml and 2286 µg/L of microcystin at station CRCC (Table 2). This high cell concentration exceeded the various threshold levels by 100's of times (Table 2), and the associated high microcystin toxin concentration of 2286 µg/L was greater than 315 times the tolerable daily intake (see TDI in Table 2 for explanation). These data illustrated that although cell density was low (2492 cells/ml) at the open water station CR01 on July 13th (Table 2), localized heavy blooms can still occur due to MSAE buoyancy and the concentrating effect of wind. Station IR01showed a relatively low level of 13,777 cells/ml and MSAE was not detected upstream of the reservoirs at KRAC or downstream of Iron Gate Dam at station KRBI (Table 2).

By the next sample period of July 26-27, data clearly showed that blooms of MSAE and associated microcystin toxin had increased substantially in intensity and extent since the July 13th sample period (Table 2; Figures 2 and 3). All reservoir stations on July 27th exceeded the

MPAHEL of 100,000 cells/ml (Figure 3; top panel). In fact, the maximum MSAE cell count of over 393 million cells/ml at CRCC exceeded the WHO MPAHEL by over 3,900 times and the MSAE specific level of 40,000 cells/ml by 9835 times (Table 2). This is the highest cell density yet recorded for the reservoirs. The open-water stations CR01 and IR01 also exceeded the MPAHEL level by 163 and 65 times, respectively. Toxin concentrations were also well above both threshold levels (Figure 3; bottom panel), with maximum toxin concentrations higher than maxima measured in 2005. Both shoreline and open-water stations exceeded the World Health Organization guideline of 20 µg/L (of microcystin) for moderate probability of adverse health effects, with levels exceeding the WHO guideline by as much as 140.7 times at station CRCC on 7/27 where a value of 2813 µg/L (2.8 mg/L) was reported. A 40 lb child accidentally ingesting 100 mls at this station would exceed the TDI by over 388 times.

Although no MSAE was detected at KRAC above Copco Reservoir on 7-26, 35,985 cells/ml were detected at KRBI below Iron Gate Reservoir; MSAE was not detected further downstream at either Seiad Valley (SV) or Orleans (OR) on 7-26 (Table 2; Figure 4). KRBI had lower microcystin (3.4 µg/L) than the recreational guidelines of 8 and 20 µg/L; however, the level was 3.4 times higher than the drinking water guideline of 1 µg/L. Despite the lack of MSAE detected at Orleans (OR), microcystin was detected at a level of 0.97 µg/L.

Overall MSAE cell density on Aug 8th was lower than July 27th; however, all reservoir stations continued to exceed the MPAHEL of 100,000 cells/ml (Figure 3). Again, MSAE was not detected at KRAC above Copco Reservoir, but 24,929 cells/ml were detected at KRBI below Iron Gate Reservoir, and due to an omission no cell density samples (see below for toxin results) were collected at either Seiad Valley (SV) or Orleans (OR). All shoreline and open-water stations exceeded the MPAHEL threshold of 20 µg/L (of microcystin). Although the maximum MSAE cell count of over 393 million cells/ml at CRCC on Jul 27th declined to a maximum of ~26.5 million cells/ml on Aug 8th, microcystin toxin level actually increased over 4x to 12,176 µg/L (or 12.18 mg/L; Table 2 and Figure 3). The trend of variable toxin production per unit cell count was also demonstrated during the 2005 sampling season; where although there was a general trend of increasing toxin with increasing cell density in 2005, there was considerable variability in the toxin to unit cell density ratio (Kann and Corum 2006). The microcystin levels of 3779 µg/L at CRMC and 12,176 µg/L at CRCC are the highest yet recorded in these reservoirs and are among the highest recorded in the world.

Despite the non-detect for MSAE cells at Klamath River Station KRAC (above Copco Reservoir) on 8/7, microcystin toxin was measured at a level of 2.0 µg/L (Table 2; Figure 4). Although no MSAE cells were detected at KRAC, cell densities of ~21,000 to 24,000 cells per ml were measured on Aug 7th in samples collected by USBR upstream from KRAC in the J.C. Boyle and Keno reservoir areas (J. Cameron, written communication). This trend indicates downstream transport of toxin from these areas. However, at station KRBI downstream from Iron Gate, microcystin increased by 50% (relative to KRAC) to 3.0 µg/L on 8/7. Microcystin concentrations at Seiad Valley (SV) and Orleans (OR) were further elevated to 6.7 and 4.1 µg/L, respectively (samples for cell density were not available for these stations on 8/7). Given that blooms can recur in slower moving or backwater areas of the river, these results indicate the potential for both higher toxin concentrations and for toxin accumulation in fish tissue.

Table 2. ***Microcystis aeruginosa*** **cell density, microcystin toxin concentration, and risk exceedance for toxigenic cyanobacteria in Copco and Iron Gate Reservoirs, 2005.**

DATE	STATION NAME	DEPTH	*Microcystis aeruginosa* (cells/ml)	*Anabaena sp.*[1] (cells/ml)	Microcystin Total (µg/L)	Exceedance of WHO moderate risk level of 100,000 cells/ml *Microcystis* (x greater than 10^5 cells/ml)	Exceedance of Posting Level of 40,000 cells/ml *Microcystis* (x greater than 40,000 cells/ml)	Exceedance of WHO moderate risk level of 20 µg/L microcystin (x greater than 20 µg/L)	Exceedance of TDI of 0.04 µg/kg/day for a 40 lb (18kg) child ingesting 100 mls (x greater than TDI)
7/12/2006	KRAC	0	0	0	ns[2]	0	0		
7/13/2006	KRBI	0	0	0	ns	0	0		
7/13/2006	CR01	1	2,492	0	ns	0	0		
7/13/2006	CRCC	0	11,783,212	6,086	2286.00	118	295	114.3	315.7
7/13/2006	IR01	1	13,377	0	ns	0	0		
7/26/2006	KRAC	0	0	0	ns	0	0		
7/27/2006	CR01	0	16,340,580	0	1003.00	163	409	50.2	138.5
7/27/2006	CRCC	0	393,395,000	0	2813.00	3934	9835	140.7	388.5
7/27/2006	IR01	0	6,504,808	0	650.00	65	163	32.5	89.8
7/27/2006	IRJW	0	25,043,386	32,214	430.00	250	626	21.5	59.4
7/27/2006	KRBI	0	35,985	0	3.40	0	1	0.2	0.5
7/26/2006	SV	0	0	0	ns	0	0		
7/26/2006	OR	0	0	0	0.97	0	0	0.0	0.1
8/7/2006	KRAC	0	0	0	2.0	0.0	0.0	0.1	0.3
8/8/2006	CR01	0	2,371,806	0	507.0	23.7	59.3	25.4	70.0
8/8/2006	CRCC	0	26,487,302	0	12176.0	264.9	662.2	608.8	1681.8
8/7/2006	CRMC	0	23,575,000	0	3779.0	235.8	589.4	189.0	522.0
8/8/2006	IR01	0	1,170,405	0	87.0	11.7	29.3	4.4	12.0
8/7/2006	IRJW	0	13,717,917	0	341.0	137.2	342.9	17.1	47.1
8/7/2006	IRCC	0	1,999,113	0	113.0	20.0	50.0	5.7	15.6
8/8/2006	IR03	0	379,668	0	ns	3.8	9.5		
8/7/2006	KRBI	0	24,929	0	3.0	0.2	0.6	0.2	0.4
8/7/2006	SV	0	ns	0	6.7			0.3	0.9
8/7/2006	OR	0	ns	0	4.1			0.2	0.6
8/23/2006	KRAC	0	0	0	0.5	0.0	0.0	0.0	0.1
8/24/2006	CR01	0	92,250	0	17.6	0.9	2.3	0.9	2.4
8/23/2006	CRCC	0	16,093,579	0	3839.0	160.9	402.3	192.0	530.2

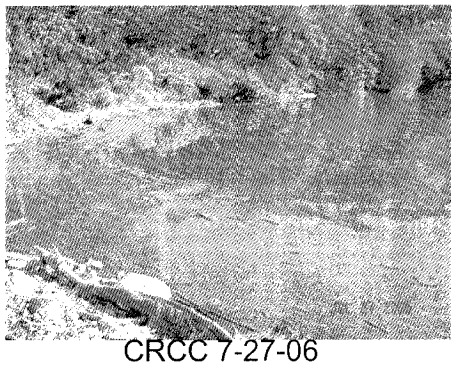
DATE	STATION NAME	DEPTH	*Microcystis aeruginosa* (cells/ml)	*Anabaena sp.*[1] (cells/ml)	Microcystin Total (µg/L)	Exceedance of WHO moderate risk level of 100,000 cells/ml *Microcystis* (x greater than 10^5 cells/ml)	Exceedance of Posting Level of 40,000 cells/ml *Microcystis* (x greater than 40,000 cells/ml)	Exceedance of WHO moderate risk level of 20 µg/L microcystin (x greater than 20 µg/L)	Exceedance of TDI of 0.04 µg/kg/day for a 40 lb (18kg) child ingesting 100 mls (x greater than TDI)
8/23/2006	CRMC	0	8,388,600	0	1543.0	83.9	209.7	77.2	213.1
8/24/2006	IR01	0	434,893	0	231.0	4.3	10.9	11.6	31.9
8/23/2006	IRJW	0	121,401	0	15.9	1.2	3.0	0.8	2.2
8/23/2006	IRCC	0	7,492,880	0	2032.0	74.9	187.3	101.6	280.7
8/23/2006	KRBI	0	28,423	0	9.2	0.3	0.7	0.5	1.3
8/23/2006	SV	0	41,299	0	7.3	0.4	1.0	0.4	1.0
8/23/2006	OR	0	31,801	0	4.6	0.3	0.8	0.2	0.6
9/6/2006	KRAC	0	0	0	BDL[3]	0.0	0.0	0.0	0.0
9/7/2006	CR01	0	95,838	0	1.4	1.0	2.4	0.1	0.2
9/7/2006	CRCC	0	3,728,535	0	206.1	37.3	93.2	10.3	28.5
9/7/2006	CRMC	0	1,028,844	0	19.0	10.3	25.7	1.0	2.6
9/7/2006	IR01	0	17,524	0	2.4	0.2	0.4	0.1	0.3
9/7/2006	IR03	0	45,585		11.9	0.5	1.1	0.6	1.6
9/7/2006	IRJW	0	11,897	0	0.7	0.1	0.3	0.0	0.1
9/7/2006	IRCC	0	29,530	0	10.1	0.3	0.7	0.5	1.4
9/6/2006	KRBI	0	3,735	0	0.7	0.0	0.1	0.0	0.1
9/6/2006	SV	0	9,555	0	0.6	0.1	0.2	0.0	0.1
9/6/2006	OR	0	3,356	0	0.4	0.0	0.1	0.0	0.1
9/20/2006	KRAC	0	0	0	BDL	0.0	0.0	0.0	0.0
9/21/2006	CR01	0	22,259	0	BDL	0.2	0.6	0.0	0.0
9/20/2006	CRCC	0	2,628,528	0	pending	26.3	65.7		
9/20/2006	CRMC	0	3,312,031	0	209.0	33.1	82.8	10.5	28.9
9/21/2006	IR01	0	12,177	0	BDL	0.1	0.3	0.0	0.0
9/20/2006	IRJW	0	8,786	0	BDL	0.1	0.2	0.0	0.0
9/20/2006	IRCC	0	8,301	0	0.19	0.1	0.2	0.0	0.0
9/20/2006	KRBI	0	3,982	0	BDL	0.0	0.1	0.0	0.0
9/20/2006	SV	0	190	0	BDL	0.0	0.0	0.0	0.0
9/20/2006	OR	0	0	0	BDL	0.0	0.0	0.0	0.0
10/4/2006	KRAC	0	0	0	pending	0.0	0.0		
10/5/2006	CR01	0	756	0	pending	0.0	0.0		

DATE	STATION NAME	DEPTH	*Microcystis aeruginosa* (cells/ml)	*Anabaena sp.*[1] (cells/ml)	Microcystin Total (µg/L)	Exceedance of WHO moderate risk level of 100,000 cells/ml *Microcystis* (x greater than 10^5 cells/ml)	Exceedance of Posting Level of 40,000 cells/ml *Microcystis* (x greater than 40,000 cells/ml)	Exceedance of WHO moderate risk level of 20 µg/L microcystin (x greater than 20 µg/L)	Exceedance of TDI of 0.04 µg/kg/day for a 40 lb (18kg) child ingesting 100 mls (x greater than TDI)
10/4/2006	CRCC	0	2,774,943	0	pending	27.7	69.4		
10/4/2006	CRMC	0	403,718	0	pending	4.0	10.1		
10/5/2006	IR01	0	0	0	pending	0.0	0.0		
10/5/2006	IR03	0	604	0	pending	0.0	0.0		
10/4/2006	IRCC	0	346	0	pending	0.0	0.0		
10/4/2006	KRBI	0	0	0	pending	0.0	0.0		
10/4/2006	SV	0	0	0	pending	0.0	0.0		
10/4/2006	OR	0	0	0	pending	0.0	0.0		
10/18/2006	KRAC	0	0	0	pending	0.0	0.0		
10/19/2006	CR01	0	313	0	pending	0.0	0.0		
10/18/2006	CR03	0	3,590,764		pending	35.9	89.8		
10/18/2006	CRCC	0	51,250,000	0	pending	512.5	1281.3		
10/18/2006	CRMC	0	10,570,313	0	pending	105.7	264.3		
10/19/2006	IR01	0	10,208	0	pending	0.1	0.3		
10/19/2006	IR03	0	0	0	pending	0.0	0.0		
10/18/2006	IRCC	0	14,455	0	pending	0.1	0.4		
10/18/2006	KRBI	0	47	0	pending	0.0	0.0		
10/18/2006	SV	0	0	0	pending	0.0	0.0		
10/18/2006	OR	0	0	0	pending	0.0	0.0		
11/1/2006	KRAC	0	0	0	pending	0.0	0.0		
11/2/2006	CR01	0	313	0	pending	0.0	0.0		
11/2/2006	CR03	0	0		pending	0.0	0.0		
11/1/2006	CRCC	0	3,855	0	pending	0.0	0.1		
11/1/2006	CRMC	0	27,234,581	0	pending	272.3	680.9		
11/1/2006	KRBI	0	0	0	pending	0.0	0.0		

[1] *Anabaena flos-aquae* (ABFA) is another potentially toxigenic cyanobacteria that can produce the neurotoxin, anatoxin-a. Because cell counts were lower than the MPAHEL of 100,000 cells/ml, ABFA is not discussed further in this report.

[2] ns=not sampled

[3] BDL= below WSU laboratory detection limit of 0.147 µg/L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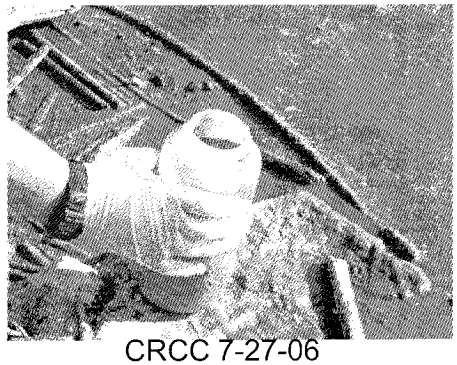
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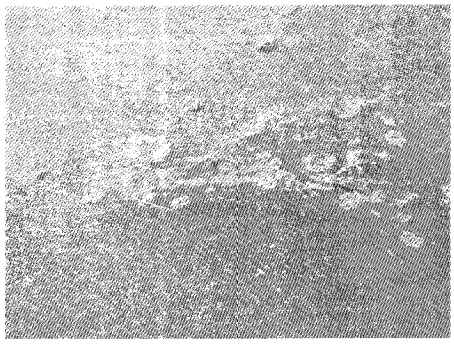
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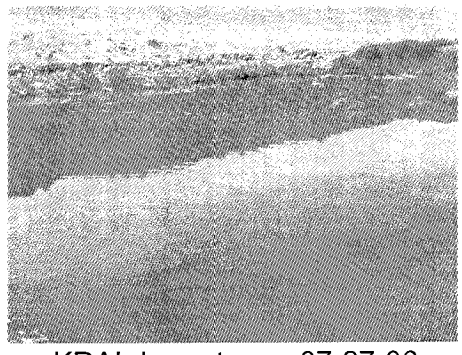
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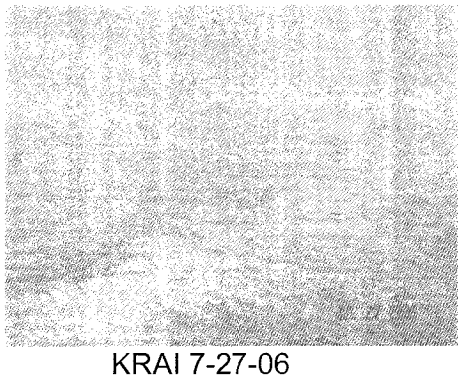
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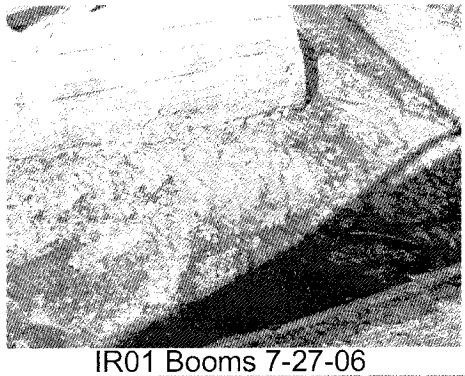
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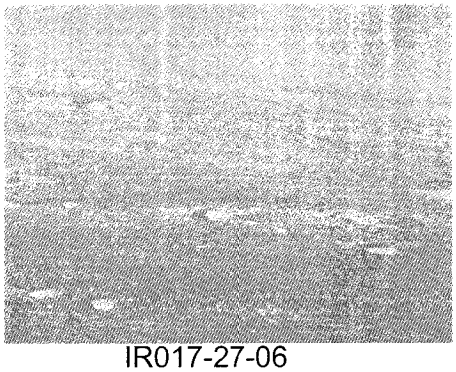
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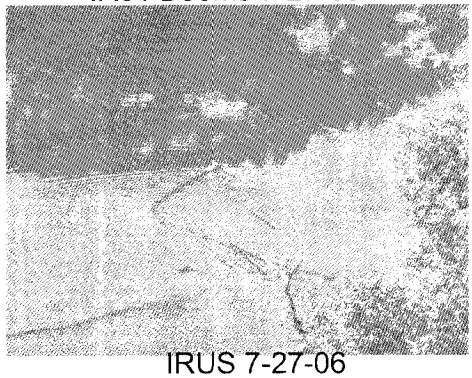
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Figure 2. *Microcystis aeruginosa* blooms in Copco and Iron Gate Reservoirs; July 26-27, 2006.

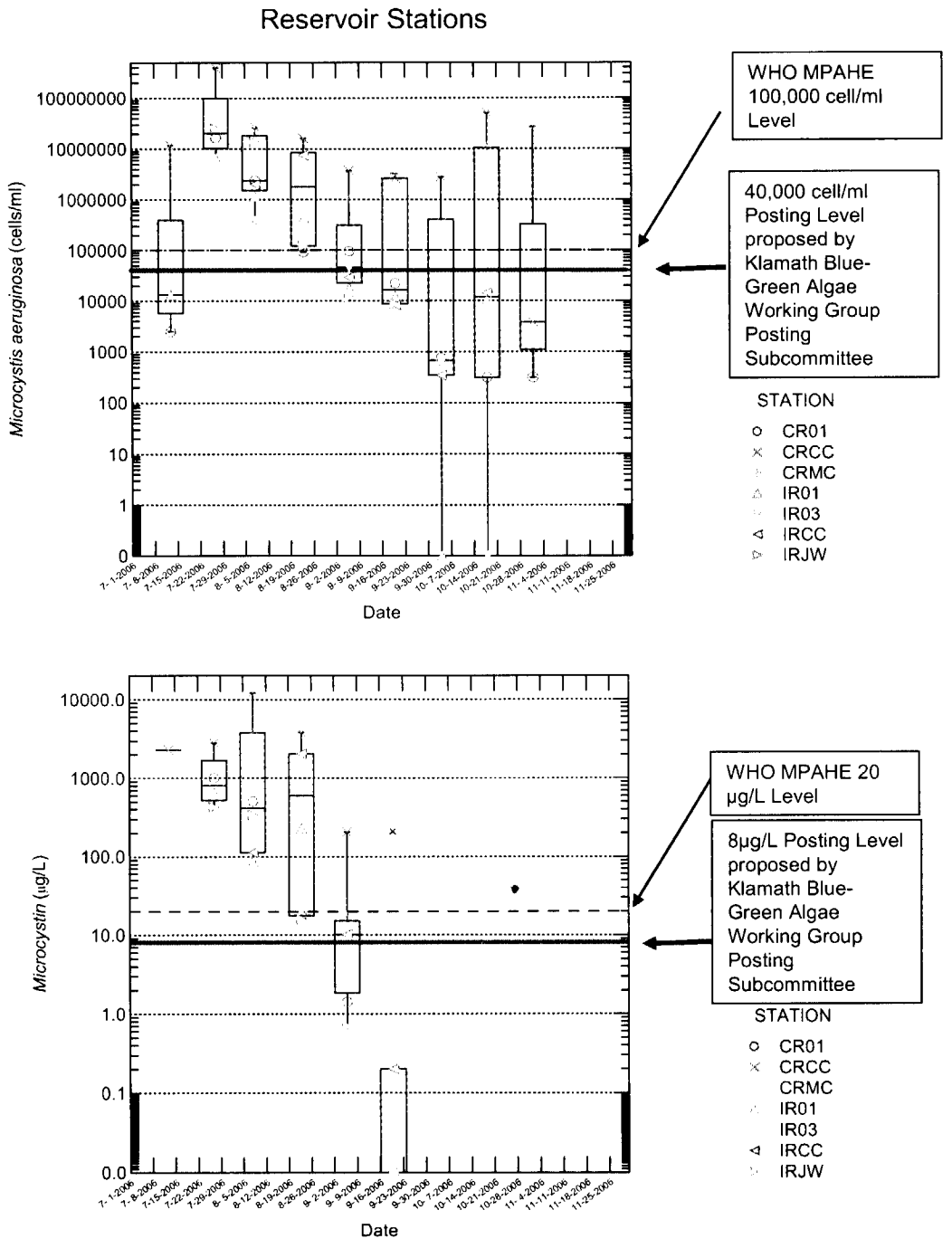

Figure 3. MSAE cell density (top) and microcystin toxin concentration (bottom) for Copco and Iron Gate Reservoir stations, 2006

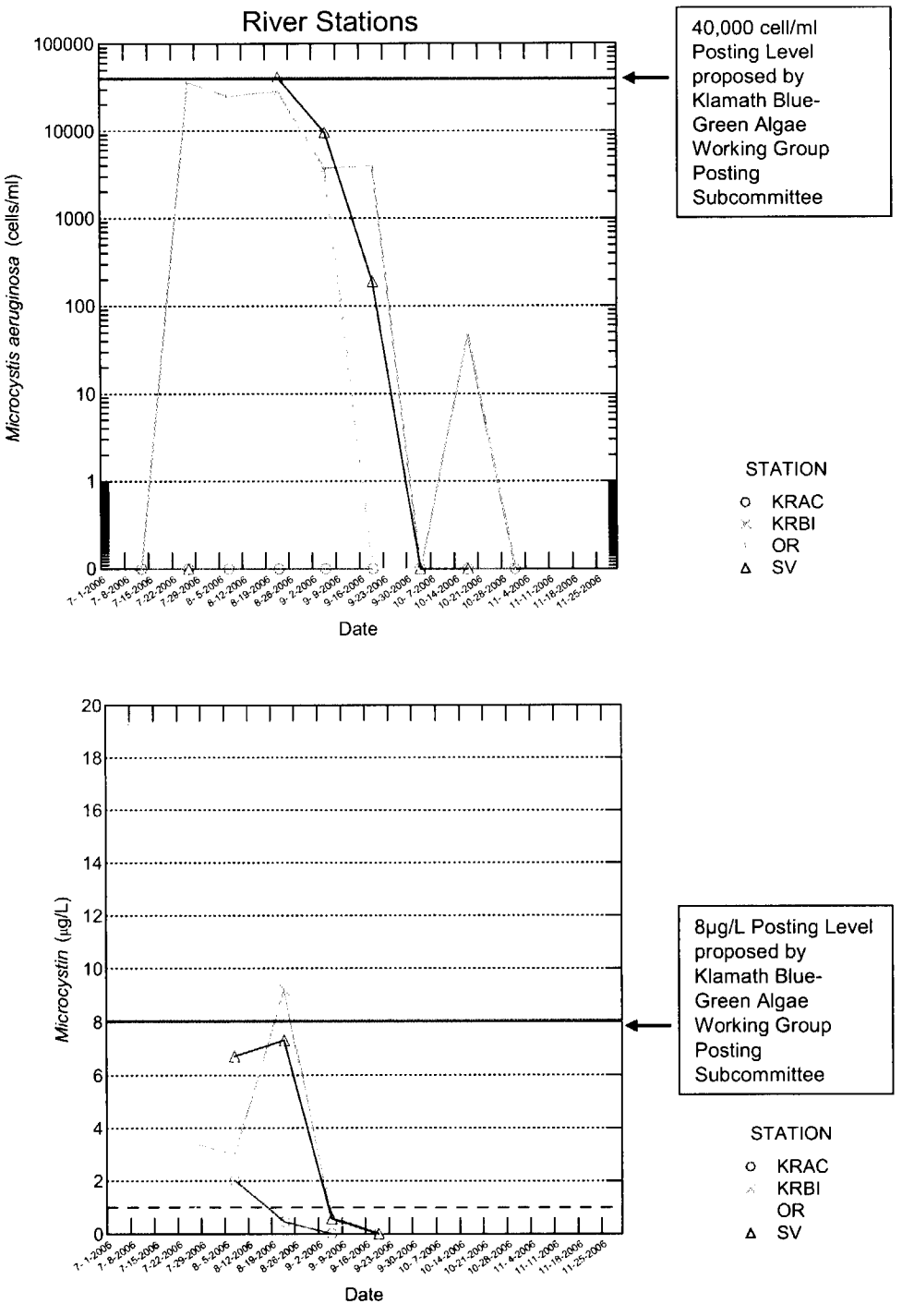

Figure 4. MSAE cell density (top) and microcystin toxin concentration (bottom) for Klamath River Stations KRAC, KRBI, SV, and OR, 2006

Although overall MSAE cell density continued to decline, all reservoir stations on August 24th were still either near or exceeded the MPAHEL for cell density and microcystin (Figure 3). Maximum exceedance in Copco Reservoir was 160.9 times the MPAHEL at CRCC (Table 2; Figure 3). Maximum Iron Gate Reservoir exceedance was 74.9 times the MPAHEL at IRCC (Table 2 Figure 3). Several stations (CRCC, CRMC, and IRCC) had >1500 µg/L (1.5mg/L) of microcystin (Table 2 and Figure 3). The microcystin levels of 3839 µg/L at CRCC and 2031 µg/L at IRCC are still among the highest recorded in these reservoirs. Similar to the previous sample period, MSAE was not detected at KRAC above Copco Reservoir, but 28,423 cells/ml were detected at KRBI below Iron Gate Reservoir, 41,299 cells/ml at Seiad Valley (SV) and 31,801 cells/ml at Orleans (OR). The Seiad Valley station exceeded the Posting Level threshold of 40,000 cells/ml (Figure 4). Again, although MSAE cells were not detected at Klamath River Station KRAC (above Copco Reservoir), microcystin toxin was measured at a low level of 0.45 µg/L (Table 2). At station KRBI downstream from Iron Gate, microcystin increased by 20 times (relative to KRAC) to 9.2 µg/L and exceeded the Posting Level of 8 µg/L (Figure 4). Concentrations at Seiad Valley (SV) and Orleans (OR) were 7.3 and 4.6 µg/L, respectively. Again these results indicate the potential for toxin accumulation in fish tissue.

Levels continued to decline in both reservoirs during September, and by September 20th only CRCC and CRMC in Copco Reservoir exceeded threshold levels for public health (Figure 3). This trend continued through the Nov 1-2nd sample period; where, with the exception of Copco Stations CRCC and CRMC, all other reservoir stations were below both the World Health Organization MPAHEL of 100,000 cells/ml and the MSAE Posting Level of 40,000 cells/ml level (Figure 3). During this same period MSAE was again not detected at KRAC above Copco, and declined to levels that were not detectable at river stations KRBI below Iron Gate Reservoir, Seiad Valley (SV), and Orleans (OR) (Table 2; Figure 4).

Microcystin levels in the reservoirs also declined though the September period, and by September 20-21 most stations were lower than 1µg/L (Table 2; Figure 3). Likewise, microcystin at all river stations dropped below 1µg/L as well (Figure 4). As noted above, October-November microcystin results are pending.

SUMMARY

Similar to 2005, the 2006 sampling program demonstrated widespread and high abundance of toxigenic MSAE blooms in Copco and Iron Gate reservoirs from July-October. MSAE cell density and toxin concentrations exceeded public health thresholds by 10 to over 1000 times during these months. Likewise, a 40 pound child accidentally ingesting 100 milliliters of reservoir water would have exceeded the WHO tolerable daily intake level by 10 to over 1600 times during dense bloom periods.

MSAE Blooms and microcystin concentrations peaked in late-July through early-August, and although values continued to exceed public health thresholds through the fall period, an overall declining trend was observed. In general, the Iron Gate decline preceded the Copco decline by several weeks. Also similar to 2005, cell density data indicated that MSAE cells were not detectable in the Klamath River directly above the reservoirs in 2006. During the period of

intense blooms in both Copco and Iron Gate reservoirs, the station above Copco (station KRAC) showed non-detects for MSAE. Conversely, the stations below Iron Gate (stations KRBI, SV, and OR), although lower in concentration than the reservoirs, followed a similar seasonal trajectory as the reservoir stations. Low concentrations of microcystin at KRAC on two occasions indicate that microcystin can be transported from upstream areas despite the lack of MSAE detection at KRAC. However, stations below Iron Gate were further elevated (by as much as 20 times on 8/23) compared to the river above Copco Reservoir.

Sample stations were intended to be representative of surface conditions at both shoreline and open-water locations. Although for the overall reservoir study samples were collected at multiple depths (e.g., 1m, 5m, 10m, 25m), the surface samples analyzed herein were specifically collected to assess recreational contact with surface water. Satellite imagery clearly shows the widespread nature of cyanobacterial blooms when they occur, and that the locations of sampling stations for this study are not unique with respect to overall spatial distribution of blooms (Figure 5).

Similar to the new Australian guideline of 50,000 cells/ml MSAE, at which point a water body is considered to be unsuitable for primary contact recreation (NHMRC 2005), WHO guidelines consider a cyanobacterial scum in a bathing (swimming) area to be cause for a high probability of adverse health effects. At that point they recommend "immediate action to control scum contact" (WHO 2003). Moreover, although the WHO MPAHEL is 20 µg/L, newly derived Australian guidelines for toxic cyanobacteria recommend 10 µg/L as a level when local authorities and health authorities should warn the public that the water body is considered to be unsuitable for primary contact recreation (NHMRC 2005). As Kann and Corum (2005) demonstrated, cell density values lower than the WHO MPAHEL of 100,000 cells/ml were associated with increased probability of exceeding critical levels. For example, consistent with the new Australian guideline of 50,000 cells/ml MSAE, the July-September 2005 model shows that microcystin values of 10 and 20 µg/L exhibited exceedance probabilities of 50% and 39% at 50,000 MSAE cells/ml (Kann and Corum 2005). These data also support the Oregon guideline levels for posting advisories that were adopted by the Posting Subcommittee of the Klamath Basin Blue-Green algae Working Group. These levels are 40,000 cells/ml of MSAE and 8 µg/L of microcystin. As noted above, these levels are specific for MSAE and microcystin, whereas the WHO levels are general levels for a variety of toxigenic cyanobacteria.

Given these existing guidelines, MSAE bloom conditions in Copco and Iron Gate Reservoirs in 2006 represented a clear public health risk with respect to water contact recreation. Maximum MSAE cell density and microcystin concentrations measured in 2006 were higher than those in 2005, and were among the highest reported in the literature (e.g., Chorus and Bartram 1999). The maximum microcystin value of 12,176 µg/L exceeded the 8 µg/L threshold level by 1522 times. Monitoring data in 2006 show that the 2005 conditions were not anomalous and that toxigenic blooms are likely to be a recurring phenomenon.

High MSAE cell density (10 to1000's of times higher than guideline levels), the presence of scums in shoreline and open-water areas, and high microcystin toxin concentrations in Copco and Iron Gate Reservoirs, necessitates prevention of primary contact recreation in, and either indirect or direct ingestion of, contaminated water in the Klamath River system.

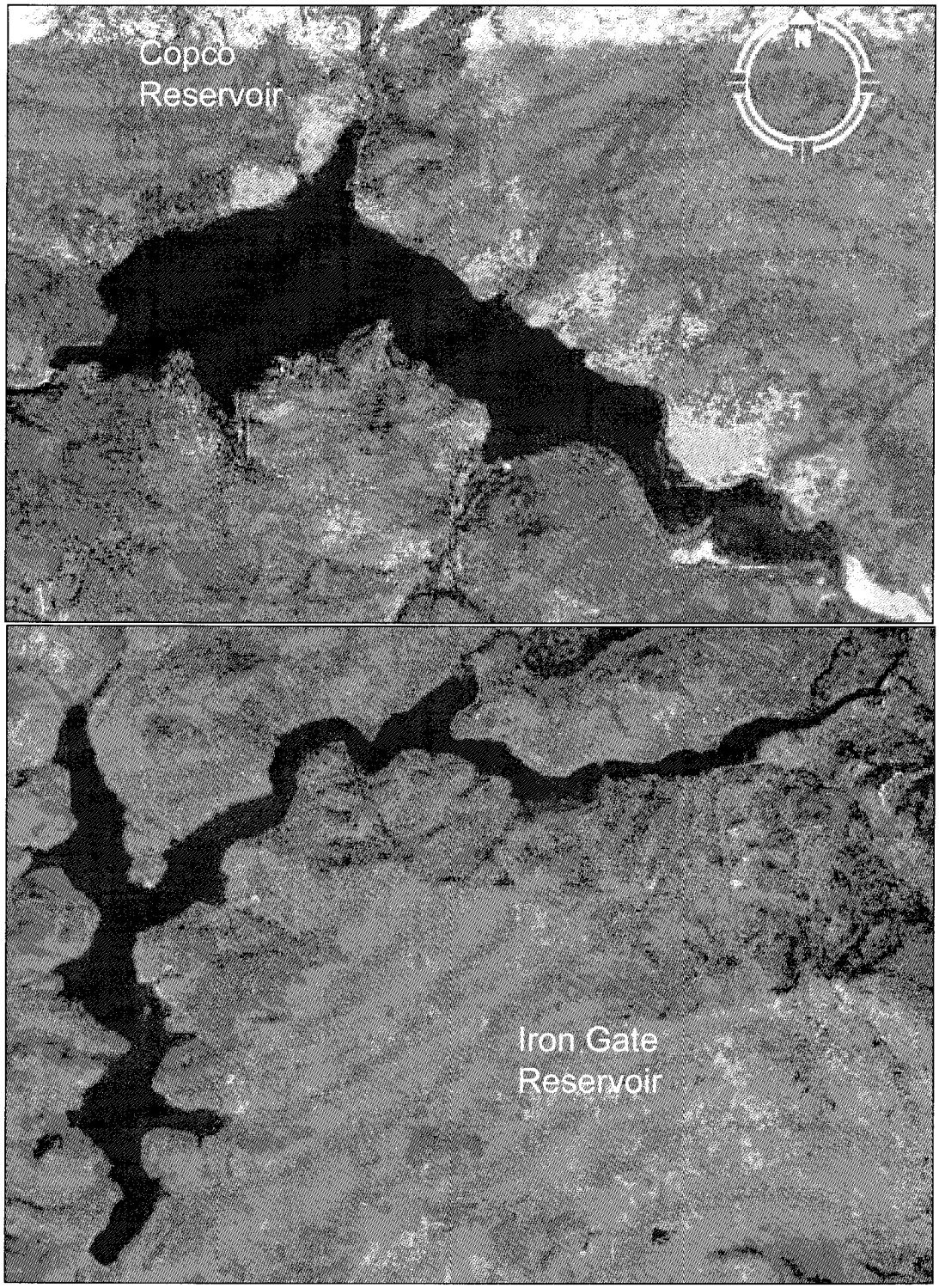

Figure 5. Satellite imagery of Copco and Iron Gate Reservoirs, CA. Image downloaded from Google Earth.

Disclaimer

Due to the patchy nature of blue-green algal blooms it is possible for higher Microcystis aeruginosa densities (and therefore higher microcystin toxin concentrations) to have been present in locations not covered in this survey, particularly along shorelines or protected coves and backwaters during calm conditions of little to no wind. Recreational users should always avoid contact with water whenever noticeable surface concentrations of algae are evident. Moreover, because pets or other domestic animals are the most likely to ingest contaminated water, these animals should not be allowed access to areas of either noticeable surface concentrations of algae or when an obvious green to blue-green appearance is evident.

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